

REGULAR ARTICLE

# Chemical composition of essential oils of *Passiflora edulis* f. *flavicarpa* agroindustrial waste

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## ABSTRACT

The study of the chemical composition of essential oils of shells and seeds of *Passiflora edulis* f. *flavicarpa* is discussed. The essential oils were obtained by microwave assisted hydrodistillation (MAHD) as extraction method using Clevenger apparatus. The compounds were separated and identified by gas chromatography mass spectrometry (GC-MS). There were no significant differences of essential oil yields between samples. 20 compounds were detected altogether. Ionol was the main compound in shells (13.15%) and seeds (12.10%). Nevertheless, some fatty acid methyl ester, aldehydes, hydrocarbons and other terpenes were found in minor quantities. These results have not been informed previously and allow adding value to this agroindustrial waste.

**Keywords:** Essential oil; Ionol; GC-MS; Residue; Yellow passion fruit

## INTRODUCTION

Yellow passion fruit (*Passiflora edulis* f. *flavicarpa*) is one of the most economically important crops of *Passiflora* species due to quality of the fruit (Zeraik et al., 2010). It is cultivated widely in tropical and semitropical regions all over the world and it is mainly used for production of concentrated juice and frozen pulp (Deng et al., 2010). Nonetheless, these processes generate a large amount of residues such as shells and seeds that represent additional operational costs and environmental issues (Leão et al., 2014; Oliveira et al., 2016).

A yellow passion fruit has approximately 58% of juice and 42% of residues altogether (FAO, 1997), whose corresponds around 34% of shells and 8% of seeds (Ayala and Cevallos, 2013). Hence, if in the first months of 2015 Ecuador exported 6,034 Tons of juice solely to Netherlands (PROEcuador, 2015), it can be inferred that approximately 4,369 tons of residues which contains shells and seeds were produced in this period.

Even though there is a great amount of generated waste as shells and seeds, some studies have shown that these

residues can be reused. Seeds have been used to extract oil by mechanical press and the seed cake obtained in this process has showed a high content of lipids with significant unsaturated fatty acids (Oliveira et al., 2016; Silva et al., 2015). In fact, fruit extracts and seed oil that contain tocopherols and phenolic compounds have been showed 2,2-diphenyl-1-picryl hydrazine (DPPH) scavenging capacity (Silva et al., 2015; Malacrida and Neuza, 2012). On the other hand, shells which are rich in fiber have been used to elaborate flour with stabilizing, emulsifying, thickening and gelling power properties similar to commercial hydrocolloid agents use in ice cream and structured fruits (Monteiro et al., 2017). Besides, they have been used to extract pectin with a high esterification degree and galacturonic acid content usefull in the food industry (De Oliveira et al., 2015).

Actually there are no studies about the chemical composition of essential oils of these residues. However, it is known that the aroma of conventional passion fruit is due to the presence of main volatile constituents as ethyl caproate and butyrate, limonene and 2-tridecanone (Dhawan et al., 2004). Therefore, the aim of this study

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was to characterize the essential oils of shells and seeds of *Passiflora edulis* f. *flavicarpa* through GC-MS in order to provide potential food applications to this agricultural waste, improve the supply chain and provide positive impact on the environment.

## MATERIALS AND METHODS

### Raw material

The raw materials used in the present study were obtained from fruits that were collected in a juice and frozen processing company in Quevedo city (Los Rios, Ecuador). The fruits were classified according to their size following the NTE INEN 1910:2012 standard. Afterward, were washed and went through by different separations processes to obtain shells and seeds prior to extraction procedure.

### Microwave assisted hydrodistillation (MAHD)

The procedure used for the extraction of essential oils of shells and seeds of *Passiflora edulis* *flavicarpa* was fitted from the method described by Rodríguez et al. (2012). Microwave assisted hydrodistillation system with Clevenger apparatus was used for extraction. Fresh raw material was distilled with water at 100°C and atmospheric pressure for four consecutive cycles of 10 minutes. The essential oils were separated with diethyl ether, were dried with anhydrous sodium sulfate, were weighed and stored in amber vials until GC-MS analysis.

### Gas chromatography mass spectrometry (GC-MS)

GC-MS analysis were carried out by a gas chromatography mass spectrometry equipment Agilent Technologies (7890A GC system and 5975C inert XL MSD with triple axis detector). A capillary column HP-5MS (30 m × 0.25 mm) with phenyl methylpolysiloxane was used as stationary phase (0.25 micron film thickness) and helium as the carrier gas (0.9 mL/min). The injection of 0.5 µL of sample (5 mg/mL for seeds and 100 mg/mL for shells) diluted in diethyl ether was done at a temperature of 250°C with splitless mode, the detector temperature was 280°C and the oven temperature was maintained at 50°C for 0.5 minutes, then it was increased to 250°C at 4°C/min. The electron ionization to 70 eV and 230°C was used as ion source and the data compounds were collect with the full scan mode (40-1000 uma) in the quadrupole mass analyzer. After all, compounds were identified by comparison of their mass spectra and mass reference of Wiley 9<sup>th</sup> with NIST 2011 MS Library.

### Statistical analysis

Yield experiments were carried out in triplicate for each sample and data were expressed as means ± standard

deviation. Differences were evaluated by the one-way analysis of variance (ANOVA), and comparison among means was determined according to Tukey test with 5% significance using InfoStat statistical software, version 2008 (Balzarini et al., 2008).

## RESULTS AND DISCUSSION

The results of MAHD and GC-MS analysis of essential oils of yellow passion fruit shells and seeds are presented below.

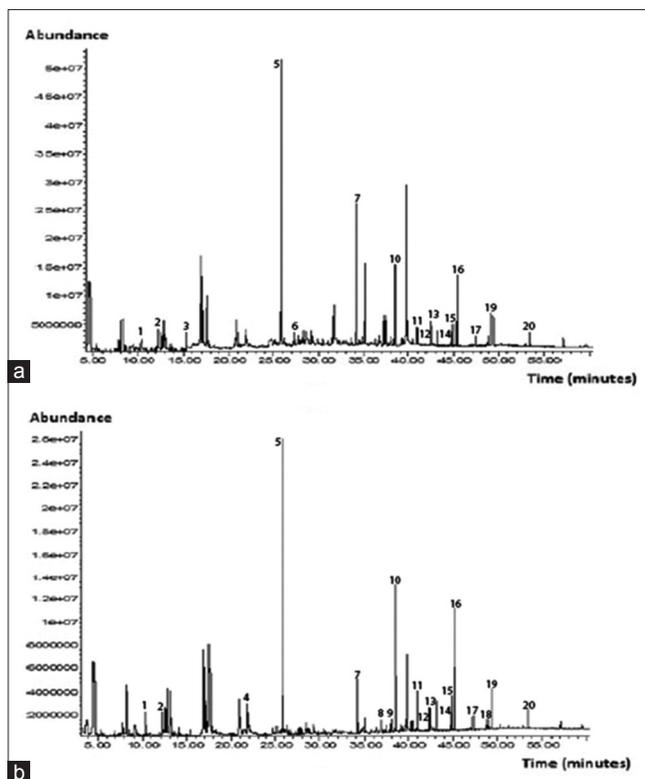
The yield percents of extracted essential oils are shown in Table 1. These results have not been referenced. The essential oil of shells (0.02222%) apparently showed to be the highest in comparison with seeds (0.01450%). Nonetheless, according to the comparative analysis there are no significant differences between samples. Probably, this is because the concentration of the samples, dispersion of the yield data obtained and the number of replicates used for each experiment. Despite the statistical outcome, these results indicate that essential oils of shells have the biggest quantity of volatile compounds. On the other hand, MAHD technique was used as extraction procedure because is superior in terms of saving energy and extraction time compared with conventional hydrodistillation. Even, GC-MS analysis of extracted essential oils indicated that the use of microwave irradiation didn't influence in the composition of the essential oils (Golmakani and Rezaei, 2008). Nonetheless, many investigations demonstrate that conventional hydrodistillation have shown higher yields than supercritical fluid extraction (SFE), microwave assisted hydrodistillation (MAHD) and solid phase micro extraction (SPME) (Omán et al., 2013). Thus, the low yield percentages obtained in this work could be explained by the extraction method employed.

The chromatograms of Fig. 1 show the chromatographic profile of essential oils of yellow passion fruit shells (a) and seeds (b). The chemical composition of the essential oils of yellow passion fruit shells and seeds are shown in Table 2, where they are listed in order of elution. Totally, 20 compounds were identified in two essential oils: 2 monoterpenes, 2 sesquiterpenes, 2 aldehydes, 6 esterified fatty acids and 8 hydrocarbons. Although most components were the same for both samples and they were formed by esterified fatty acids and hydrocarbons, the dominant

**Table 1: Yield percents of essential oils of shells and seeds of *Passiflora edulis* f. *flavicarpa***

Sample	Yield (%)
Shells	0.0222200±0.0105027 <sup>a</sup>
Seeds	0.0144967±0.0029120 <sup>a</sup>

Means values (n=3) ± standard deviation, <sup>a</sup>Different letters indicate significant differences between samples according to Tukey test (p ≤ 0.05)



**Fig 1.** Chromatographic profiles of essential oils of shells (a) and seeds (b) of *Passiflora edulis* f. *flavicarpa* obtained by GC-MS.

**Table 2: Chemical composition of essential oil of shells and seeds of *Passiflora edulis* f. *flavicarpa***

Peak	Retention time (min)	Compound	Peak area %	
			Shells	Seeds
1	10.295	Benzeneacetaldehyde	0.83	1.69
2	12.197	Linalool	1.06	0.89
3	15.335	$\alpha$ -Terpineol	0.65	ND
4	21.817	Hexyl caproate	ND	1.16
5	25.780	Ionol	13.15	12.10
6	27.251	d-Nerolidol	0.88	ND
7	34.133	Palmitaldehyde	6.24	2.18
8	36.875	Methyl Palmitate	ND	0.51
9	38.008	Ethyl 9-Hexadecenoate	ND	0.51
10	38.523	Ethyl Palmitate	3.56	5.79
11	40.941	Heneicosane	0.83	1.66
12	42.340	Ethyl Linoleate	1.00	0.98
13	42.474	Ethyl Oleate	0.94	0.98
14	43.148	Docosane	0.78	1.29
15	44.860	9-Tricosene	0.90	1.36
16	45.279	Heptadecane	3.29	5.13
17	47.315	Eicosane	0.45	0.59
18	48.925	1-Nonadecene	ND	0.41
19	49.275	Pentacosane	1.46	1.74
20	53.404	Octadecane	0.89	1.08

ND: Not detected

compound was ionol. This cyclic sesquiterpen showed the highest area percent in shells (13.15%) and seeds (12.10%). These results are similar to previous studies for essential

oils of *Passiflora ligularis* Juss, where ionol was the main compound for shells and one of the majorities in seeds (Chóez et al., 2015). Also, It was reported as a volatile compound of yellow passion fruit flavor for the first time in 1998 (Werkhoff et al., 1998). Hence, these findings prove that ionol is a distinctive compound existing in some essential oils of *Passiflora* species.

Even though the yields of essential oils are pretty low, it should be considered the high amounts of cultivated fruits and waste generated (PROECUADOR, 2015), in order to take advantage of the identified compounds. In this context, it will be presented some food and pharmacological applications of the main substances.

Ionol could be used as food additive. In fact, it was approved as flavouring agent by World Health Organization (WHO) because it wasn't showed health risks. This sesquiterpen and other related derivatives compounds weren't exceed the threshold for human intake (1800 ug per day) and they metabolized to innocuous products (WHO, 1998).

It is evident that ionol was the main compound in two samples. However, palmitaldehyde (6.24%) presented the second higher area percent in shells but no in seeds. The esterified fatty acid ethyl palmitate (5.79%) and the hydrocarbon heptadecane (5.13%) showed to be the second major compounds in seeds indeed.

Heptadecane has been identified in minor amounts in organic and conventional passion fruit pulp (Macoris et al., 2011). Palmitaldehyde was detected in shells and seeds, this fatty aldehyde has been reported previously in essential oil of *Passiflora ligularis* Juss juice and it was showed to be one of the major volatile compounds of this specie too (Chóez et al., 2015). Another aldehyde found was benzeneacetaldehyde, this compound has been reported in yellow passion fruit (Jordán et al., 2000) and it is known as a key odor compound that plays a central role in pollinator attraction and floral isolation (Falcão et al., 2008).

On the other hand, the esterified fatty acid ethyl palmitate has been used as flavoring ingredient in foods and it is listed in the Encyclopedia of Food and Color Additives indeed (Burdock, 1997). Also, ethyl 9-hexadecenoate has been reported as additive for alcoholic beverages, specifically in the whiskey industry (Nykänen and Suomalainen, 1983) and methyl palmitate has been demonstrated anti-inflammatory and antifibrotic effects in experimental animal models of liver injury (Cai et al. 2005; Rodriguez-Rivera et al. 2008). Furthermore, methyl palmitate has been combated inflammation in several models of inflammatory disorders such as: paw edema, lipopolysaccharide-induced endotoxemia, lipopolysaccharide-induced acute lung injury

and croton oil-induced ear edema in rats (Saeed et al. 2012; Sharawy et al. 2013). Moreover, is one of the best known ligands for pheromone receptors Or47b and Or88a (Dweck et al. 2015). This last compound was detected only in seeds. These results coincide with the finding of essential oils of *Passiflora ligularis* Juss where methyl palmitate showed to be present as a volatile constituent of seed essential oil of this *Passiflora* specie. Neither in shells nor juice it wasn't detected (Chóez et al., 2015). In addition, ethyl 9-hexadecenoate was detected just in seeds too. On the contrary, it has been detected in aerial parts of *Passiflora incarnate* L. methanolic extract (Aman et al., 2016).

Equally important, hexil caproate (1.16%) was detected in minor quantities just in seed essential oil. This fatty ester has been described as the majority volatile compound of passion fruit juice (Hiu and Scheuer, 1961) and it is used in fragrances of soap, detergent, cream, lotion and perfumes. Additionally, it is utilized as artificial flavoring of foodstuffs and it has been approved by the Food and Drug Administration (FDA) for food use (Opdyke, 1978).

Alternatively, the high presence of aliphatic compounds with minor peak area percents such as heneicosane, docosane, 9-tricosene, eicosane, 1-nonadecene, pentacosane and octadecane could be explained by the gums and/or resins of vegetal matrixes. These compounds are freed from such products by distillation (Hüsni et al., 2007). The results indicate that peak area of esterified fatty acids with the exception of ethyl linoleate were higher in seeds. In fact, the three esterified fatty acids that elute first weren't identified in shells. In the same way, the long-chain alkanes have been showed a similar relation but 1-nonadecene wasn't detected in shells.

Instead of the low presence of monoterpenes and sesquiterpenes in both samples, linalool was identified in shells (1.06%) and seeds (0.65%) but  $\alpha$ -terpineol (0.65%) and d-nerolidol (0.88%) were detected only in shells. So, it can be inferred that these last compounds are characteristics of this residue. Linalool has been described in tropical fruits such as passion fruit and banana (Jordán et al., 2000). Even it has been reported as one of main compounds of essential oil of *Passiflora incarnate* L (Buchbauer and Jirovetz, 1992) and it has been informed as molluscicidal, larvicidal (Yang et al., 2014), analgesic, anti-inflammatory and antioxidant (Seol et al., 2016). Equally important  $\alpha$ -terpineol has been described as one of the major flavor constituents of yellow passion fruit (Hui et al., 2010), this terpene is responsible of the floral aroma of passion fruit and it is considered as a marker volatile compound (Janzanti and Monteiro, 2014; Conde-Martínez et al, 2014). Further, it has reported significant biological activities such as antimicrobial, antispasmodic, anticonvulsant and

antinociceptive immunostimulant (Oliveira et al., 2016). It is considerable to mention that linalool and  $\alpha$ -terpineol have been described before as aroma compounds for organic passion fruit. In fact, the abundance of these terpene alcohols increased in the ripeness process of the pulp (Janzanti and Monteiro, 2014).

## CONCLUSIONS

These findings suggest that essential oils of *Passiflora edulis* f. *flavicarpa* shells and seeds might be useful as raw material for food additive due to the properties previously reported of the main constituents ionol and ethyl palmitate. These two essential oils could be used as flavor and aroma substances in the food industry. Additionally, in accordance to outcomes the cyclic sesquiterpen ionol showed to be a major volatile compound of these agroindustrial waste essential oils.

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### Author contributions

I. C. G. and M. M. M. designed, conducted the work and wrote the paper, A. O. made the experimental work and P. I. M. S. made a major contribution to the review paper.

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